

Original citation:

Agoti, Charles N., Kiyuka, Patience K., Kamau, Everlyn, Munywoki, Patrick K., Bett, Anne, van der Hoek, Lia, Kellam, Paul, Nokes, D. James and Cotten, Matthew. (2016) Human Rhinovirus B and C genomes from rural coastal Kenya. *Genome Announcements*, 4 (4). e00751.

Permanent WRAP URL:

<http://wrap.warwick.ac.uk/80591>

Copyright and reuse:

The Warwick Research Archive Portal (WRAP) makes this work of researchers of the University of Warwick available open access under the following conditions.

This article is made available under the Creative Commons Attribution 4.0 International license (CC BY 4.0) and may be reused according to the conditions of the license. For more details see: <http://creativecommons.org/licenses/by/4.0/>

A note on versions:

The version presented in WRAP is the published version, or, version of record, and may be cited as it appears here.

For more information, please contact the WRAP Team at: wrap@warwick.ac.uk

Original citation:

Agoti, Charles N., Kiyuka, Patience K., Kamau, Everlyn, Munywoki, Patrick K., Bett, Anne, van der Hoek, Lia, Kellam, Paul, Nokes, D. James and Cotten, Matthew. (2016) Human Rhinovirus B and C genomes from rural coastal Kenya. *Genome Announcements*, 4 (4). e00751.

Permanent WRAP URL:

<http://wrap.warwick.ac.uk/80591>

Copyright and reuse:

The Warwick Research Archive Portal (WRAP) makes this work of researchers of the University of Warwick available open access under the following conditions.

This article is made available under the Creative Commons Attribution 4.0 International license (CC BY 4.0) and may be reused according to the conditions of the license. For more details see: <http://creativecommons.org/licenses/by/4.0/>

A note on versions:

The version presented in WRAP is the published version, or, version of record, and may be cited as it appears here.

For more information, please contact the WRAP Team at: wrap@warwick.ac.uk

Human Rhinovirus B and C Genomes from Rural Coastal Kenya

Charles N. Agoti,^{a,b} Patience K. Kiyuka,^a Evelyn Kamau,^a Patrick K. Munywoki,^{a,b} Anne Bett,^a Lia van der Hoek,^c Paul Kellam,^{d,e} D. James Nokes,^{a,f} Matthew Cotten^g

Epidemiology and Demography Department, KEMRI—Wellcome Trust Research Programme, Kilifi, Kenya^a; School of Health and Human Sciences, Pwani University, Kilifi, Kenya^b; Laboratory of Experimental Virology, Academic Medical Center of the University of Amsterdam, Amsterdam, The Netherlands^c; Imperial College London, London, United Kingdom^d; KyMab Limited, Cambridge, United Kingdom^e; School of Life Sciences and WIDER, University of Warwick, Coventry, United Kingdom^f; The Wellcome Trust Sanger Institute, Cambridge, United Kingdom^g

Primer-independent agnostic deep sequencing was used to generate three human rhinovirus (HRV) B genomes and one HRV C genome from samples collected in a household respiratory survey in rural coastal Kenya. The study provides the first rhinovirus genomes from Kenya and will help improve the sensitivity of local molecular diagnostics.

Received 6 June 2016 Accepted 10 June 2016 Published 28 July 2016

Citation Agoti CN, Kiyuka PK, Kamau E, Munywoki PK, Bett A, van der Hoek L, Kellam P, Nokes DJ, Cotten M. 2016. Human rhinovirus B and C genomes from rural coastal Kenya. *Genome Announc* 4(4):e00751-16. doi:10.1128/genomeA.00751-16.

Copyright © 2016 Agoti et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Matthew Cotten, mc13@sanger.ac.uk.

Human rhinoviruses (HRVs) are nonenveloped viruses with a single-stranded positive-sense RNA genome of approximately 7,200 nucleotides with a single open reading frame (ORF) encoding a polyprotein that is cleaved into four structural proteins and seven nonstructural proteins (1). These viruses belong to the *Enterovirus* genus within the *Picornaviridae* family, with three species currently defined (HRV-A, HRV-B, and HRV-C) (2). In addition to being widely recognized as the major cause of common cold syndrome, HRVs have been increasingly diagnosed in individuals with lower respiratory tract infections (3), although underlying causality remains uncertain (3–5).

Typical of RNA viruses, HRVs can evolve rapidly due to an error-prone RNA-dependent RNA polymerase, short-generation times, and large population sizes. To maintain the sensitivity of clinical molecular diagnostics for these viruses, it is essential that diagnostic primers recognize currently circulating viruses. It is therefore useful to routinely sequence a subset of clinical samples using unbiased, random priming methods combined with *de novo* assembly genome methods (6). The resulting sequence data provide a less biased description of the circulating viruses in a community, and the approach has the potential to reveal viruses that diagnostic primers fail to detect. As part of a household cohort study undertaken in Kilifi county, coastal Kenya, samples were collected from every household member twice weekly, independent of symptoms (7). As a by-product of an agnostic sequencing of coronavirus positive samples detected in the survey, we generated four nearly complete rhinovirus genomes, three HRV-B and one HRV-C, which are reported here. These data represent the first HRV full genomes from Kenya.

Phylogenetic analysis of the VP4-2 junctions of each viral genome compared with reference sequences revealed that the HRV-B viruses belonged to type HRV-B37 ($n = 1$) and HRV-B48 ($n = 2$), while the single HRV-C genome was assigned to HRV-C35 (2). The two HRV-B48 viruses were identical in the VP4-2 region, but four nucleotide differences were noted elsewhere in their genomes.

Of interest, the real-time reverse transcriptase PCR primers used in the local viral diagnostic lab (8) were found to have nucleotide differences from the HRV-B genomes reported here. Several mismatches to both the forward and reverse primers are predicted to compromise PCR function. In fact, the three samples yielding HRV-B genomes were rhinovirus-negative by the diagnostic assay. It is noteworthy that HRV-B was identified at very low prevalence in this household cohort (unpublished data) and the samples yielding HRV-B genomes were classified as negative by this PCR assay. While it was previously reported that HRV-B circulates at lower prevalence than either HRV-A or C (3, 9), it cannot be excluded that a contributing factor to the low HRV-B prevalence was the diagnostic assay failing to detect some of the locally circulating HRV strains. The availability of local full genomes generated using a primer-independent method provides an important tool for improving diagnostic methods.

Nucleotide sequence accession numbers. The complete genome sequences of the rhinovirus genomes are available at GenBank under the accession numbers [KX348029](https://www.ncbi.nlm.nih.gov/nuccore/KX348029) to [KX348032](https://www.ncbi.nlm.nih.gov/nuccore/KX348032).

ACKNOWLEDGMENT

This work was published with the permission of the Director of KEMRI.

FUNDING INFORMATION

This work was funded by the Wellcome Trust (grants 093724 and 102975).

REFERENCES

- Jacobs SE, Lamson DM, St George K, Walsh TJ. 2013. Human rhinoviruses. *Clin Microbiol Rev* 26:135–162. <http://dx.doi.org/10.1128/CMR.00077-12>.
- McIntyre CL, Knowles NJ, Simmonds P. 2013. Proposals for the classification of human rhinovirus species A, B and C into genotypically assigned types. *J Gen Virol* 94:1791–1806. <http://dx.doi.org/10.1099/vir.0.053686-0>.
- Onyango CO, Welch SR, Munywoki PK, Agoti CN, Bett A, Ngama M, Myers R, Cane PA, Nokes DJ. 2012. Molecular epidemiology of human rhinovirus infections in Kilifi, coastal Kenya. *J Med Virol* 84:823–831. <http://dx.doi.org/10.1002/jmv.23251>.

4. Berkley JA, Munywoki P, Ngama M, Kazungu S, Abwao J, Bett A, Lassaunière R, Kresfelder T, Cane PA, Venter M, Scott JA, Nokes DJ. 2010. Viral etiology of severe pneumonia among Kenyan infants and children. *JAMA* 303:2051–2057. <http://dx.doi.org/10.1001/jama.2010.675>.
5. Pretorius MA, Tempia S, Walaza S, Cohen AL, Moyes J, Variava E, Dawood H, Seleka M, Hellferscee O, Treurnicht F, Cohen C, Venter M. 2016. The role of influenza, RSV and other common respiratory viruses in severe acute respiratory infections and influenza-like illness in a population with a high HIV sero-prevalence, South Africa 2012–2015. *J Clin Virol* 75:21–26. <http://dx.doi.org/10.1016/j.jcv.2015.12.004>.
6. Cotten M, Oude Munnink B, Canuti M, Deijs M, Watson SJ, Kellam P, van der Hoek L. 2014. Full genome virus detection in fecal samples using sensitive nucleic acid preparation, deep sequencing, and a novel iterative sequence classification algorithm. *PLoS One* 9:e93269. <http://dx.doi.org/10.1371/journal.pone.0093269>.
7. Munywoki PK, Koech DC, Agoti CN, Lewa C, Cane PA, Medley GF, Nokes DJ. 2014. The source of respiratory syncytial virus infection in infants: a household cohort study in rural Kenya. *J Infect Dis* 209: 1685–1692. <http://dx.doi.org/10.1093/infdis/jit828>.
8. Gunson RN, Collins TC, Carman WF. 2005. Real-time RT-PCR detection of 12 respiratory viral infections in four triplex reactions. *J Clin Virol* 33: 341–344. <http://dx.doi.org/10.1016/j.jcv.2004.11.025>.
9. Smuts HE, Workman LJ, Zar HJ. 2011. Human rhinovirus infection in young African children with acute wheezing. *BMC Infect Dis* 11:65. <http://dx.doi.org/10.1186/1471-2334-11-65>.